

BIONETICS

MUTAGENIC EVALUATION OF

COMPOUND 000299296

FERROUS GLUCONATE

(1-63)

5516 Nicholson Lane Kensington, Maryland 20795 MUTAGENIC EVALUATION OF

COMPOUND 000299296

FERROUS GLUCONATE

(1-63)

SUBMITTED TO

FOOD & DRUG ADMINISTRATION
DEPARTMENT OF HEALTH, EDUCATION AND WELFARE
ROCKVILLE, MARYLAND

SUBMITTED BY

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APRIL 30, 1975



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EVALUATION SUMMARY

Compound 000299296, Ferrous Gluconate, appeared to be mutagenic for indicator strain TA-1538 in activation tests with primate liver activation homogenate.



DATE:

04/30/75

SPONSOR:

Food and Drug Administration, Contract Number 223-74-2104

SUBJECT: Evaluation of Test Compound 000299296, Ferrous Gluconate

I. **OBJECTIVE**

The objective of this study was to evaluate the test compound for genetic activity in microbial assays with and without the addition of mammalian metabolic activation preparations.

II. MATERIALS

Α. Test Compound

Date Received: 1.

August, 1974

2.

Description: Fine grey powder

В. Indicator Microorganisms

The following strains of indicator microorganisms were used in the evaluation:

Yeast Strain:

Saccharomyces cerevisiae, strain D4

Bacteria Strains: Salmonella typhimurium, strains:TA-1535

TA-1537

TA-1538

C. Reaction Mixture

The following reaction mixture was employed in the activation tests:

Component	Final Concentration/ml
 TPN (sodium salt) Isocitric acid Tris buffer, pH 7.4 MgCl₂ Tissue homogenate fraction 	6 μ M 49 μ M 28 μ M 1.7 μ M 72 mg



D. <u>Tissue Homogenates and Supernatant</u>

The tissue homogenates and 9,000 x \underline{g} supernatants were prepared from tissues of the following mammalian species: Mouse-ICR random bred adult males; rat-Sprague-Dawley adult males; and primate-Macaca mulatta adult males.

E. Positive Control Compounds

Table 1 lists chemicals for positive controls in the direct and activation assays.

TABLE 1 POSITIVE CONTROLS USED IN DIRECT AND ACTIVATION ASSAYS

<u>Assay</u>	<u>Chemical^a</u>	Solvent	Probable Mutagenic Specificity
Non-activation	Ethyl methanesulfonate	Water or saline	BPS
	2-Nitrofluorene	Dimethylsulfoxide ^C	FS
	Quinacrine mustard	Water or saline	FS
Activation	Dimethylnitrosamine	Water or saline	BPS
	2-Acetylaminofluorene	Dimethylsulfoxide ^C	FS

a Concentrations given in the Results Section

Previously shown to be non-mutagenic

III. METHODS

A. Toxicity

The solubility, toxicity and doses for all chemicals were determined prior to screening.

Each chemical was tested for survival against the specific indicator strains over a range of doses to determine the 50% survival dose. Bacteria were tested in phosphate buffer, pH 7.4, for one hour at 37°C on a shaker. Yeasts were tested in phosphate buffer, pH 7.4, for four hours at 30°C on a shaker. The 50% survival curve and the 1/4 and 1/2 50% doses calculated.

If no toxicity was obtained for a chemical with a given strain, then a maximum dose of 5% (w/v) was used against the strain.

Unless otherwise specified, the doses calculated for the tests in buffer were applied to the activation tests. The solubility of the test chemical under treatment conditions is stated in the Results Section.



c BPS = base-pair substitution; FS = frameshift

B. Plate Tests

In the nonactivation procedure, approximately 10° cells of a log-phase culture of the bacterial indicator strains were spread over the surface of a minimal plate, and a measured amount of the test chemical was placed in the center of the test plate. In activation tests, the test chemical was added to the cells, and an aliquot of the mixture was spread on the surface of the test plate. The reaction mixture (0.1 ml) plus tissue extract was then spotted on the surface of the plate. Positive and solvent controls were included. All plates were incubated at 37°C for four days and then scored. Each compound (test, positive control and solvent control) was done in duplicate. Concentrations of the positive control compounds are listed in the Results Section.

C. <u>Suspension Tests</u>

Non activation

Log-phase bacteria and stationary-phase yeast cultures of the indicator organisms were grown in complete broth, washed and resuspended in 0.9% saline to densities of 1 x 10^9 cells/ml and 5 x 10^7 cells/ml, respectively. This constituted the working stock for tests of a group of test chemicals and their respective controls. Tests were conducted in plastic tissue culture plates. Cells plus appropriate volume(s) of the test chemical were added to the wells to give a final volume of 1.5 ml. The solvent replaced the test chemical in the negative controls. Treatment was at 30°C for four hours for yeast tests and at 37°C for one hour for bacterial tests. All flasks were shaken during treatment. Following treatment, the plates were set on ice. Aliquots of cells were removed, diluted in sterile saline (4°C) and plated on the appropriate complete media. Undiluted samples from flasks containing the bacteria were plated on minimal selective medium in reversion experiments. Samples from a 10^{-1} dilution of treated cells were plated on the selected media for enumeration of gene conversion with strain D4. Bacterial plates were scored after incubation for 48 hours at 37°C. The yeast plates were incubated at 30°C for 3-5 days before scoring.

Activation

Bacteria and yeast cells were grown and prepared as described in the non activation tests. Measured amounts of the test and control chemicals plus 0.25 ml of the stock-cell suspension were added to wells of the Linbro plate containing the appropriate tissue fraction and reaction mixture. All flasks (bacteria and yeast) were incubated at 37°C in an oxygen atmosphere with shaking. The treatment times as well as the dilutions, plating procedures and scoring of the plates were the same as described for non activation tests.



D. Preparation of Tissue Homogenates and 9,000 x g Cell Fractions

Male animals (sufficient to provide the necessary quantities tissues) were killed by cranial blow, decapitated and bled. Organs were immediately dissected from the animal using aseptic techniques and placed in ice-cold 0.25 M sucrose buffered with Tris at pH of 7.4. Upon collection of the desired quantity of organs, they were washed twice with fresh buffered sucrose and completely homogenized with a motor-driven homogenizing unit at 4° C. The whole organ homogenate obtained from this step was divided into two samples. One sample was frozen at -80° C and the other was centrifuged for 20 minutes at 9,000 x g in a refrigerated centrifuge. The supernatant from the centrifuged sample was retained and frozen at -80° C. These two frozen samples were used for the activation studies.

E. Data Recording and Reporting

Following the specified incubation periods all population plates were scored by an automatic colony counter and the results from each plate of a set were recorded, in ink, on data processing forms. All minimal or other types of selective media plates were hand scored and the results recorded along with the respective population data. Other relevant experimental data were recorded on experimental definition forms. For bacteria strains the number of colonies recorded from either the population or selective plates represents that number in 1 ml of test suspension plated. The numbers recorded for the yeast strain D4 represent the number in 0.5 ml of test suspension plated. Data was then processed and printed from a computer program.



- IV. RESULTS SECTION
- A. Solubility Properties of the Test Compound
- Name or code designation of the test compound: 000299296, Ferrous Gluconate
- 2. Test solvent: Saline
- 3. Solubility of the test compound under treatment conditions: Soluble under treatment conditions.
- 4. Additional comments: Fine grey powder
- B. Toxicity and Dosage Determinations for the Test Compound 000299296
- 1. Test date for toxicity determination: 01/28/75
- 2. The 50% survival level was determined for bacteria and yeast indicator organisms by conducting survival curves with the test compound at the following concentrations:

Percent Concentration (w/v or v/v)

5.0 0.5 0.05 0.005 0.0005

3. Concentrations of the test compound used in the mutagenicity tests:

	Percent Concentration		
Dose	Bacteria	Yeast	
1/4 50% Survival	0.035	2.5	
1/2 50% Survival	0.07	5.0	
50% Survival	0.14	10.0	
Plate Tests	0.07		



IV. SUMMARY OF TEST RESULTS

Plate Tests

A. Name or code designation of the test compound: 000299296

B. Test date: 02/14/75

C. Concentration of the test compound: 0.07%

<u>Test</u>		Species	Tissue	<u> </u>	A-1535		<u>-1537</u>		<u>-1538</u>
1.	Non-activation				2	1	2	1	2
	Solvent Control Positive Control ^a Test Compound			47 >10 47	78 ³ >10 ³ 17	11 209 3	14 301 3	26 147 11	6 140 6
2.	<u>Activation</u>								
	Negative Control Solvent Control Reaction Mixture			40 27	90 89	14 10	22 33	14 17	4 18
	Control			47	31	12	34	20	22
	Positive Control ^b Positive Control Positive Control	Mouse	Liver Lung Testes	59	>500 59 60	83 10 11	75 7 10	>200 20 11	>200 10 10
	Positive Control Positive Control Positive Control	Rat	Liver Lung Testes	63	>300 77 61	83 10 10	86 7 8	>10 ² 19 12	>10 ² 9 9
	Positive Control Positive Control Positive Control	Monkey	Liver Lung Testes	71	>100 54 61	41 9 10	34 8 9	>100 20 13	93 6 10
	Test Compound Test Compound Test Compound	Mouse	Liver Lung Testes	16 35 35	23 15 25	7 0 2	3 4 0	2 1 2	2 2 3
	Test Compound Test Compound Test Compound	Rat	Liver Lung Testes	20 33 s 32	19 17 26	4 0 2	3 3 1	2 2 3	2 2 3
	Test Compound Test Compound Test Compound	Monkey	Liver Lung Testes	16 35 s 29	31 23 26	4 0 2	1 6 1	3 1 2	2 3 4
a	TA-1537 QM 20	μl/plate μg/plate μg/plate	7	TA-1535 TA-1537 TA-1538	AAF	100	µm/pla µg/pla µg/pla	ite	



DATA TABLE TERMS AND ABBREVIATIONS

OR TERM	DEFINITION OR EXPLANATION
COMPOUND	Client designated compound number appears in this column.
TEST CODES	NAN = Non Activation: Solvent Control NAP = Non Activation: Positive Control NA1 = Non Activation: Test Compound Dose 1 NA2, etc. = Reflects the other dose level(s)
	A+C = Negative Chemical Control A-C = Activation: Solvent Control ACP = Activation: Positive Control ACT = Activation: Test Compound A+T = Activation: Tissue Control
	LI = Liver Tissue Activation Fraction LU = Lung Tissue Activation Fraction KI = Kidney Tissue Activation Fraction TE = Testes Tissue Activation Fraction 1,2, etc. = Dose Levels
CONCENTRATION	All test compound dose levels are expressed as a whole number followed by an exponent (negative) identified by the appropriate units.
	Example: 0025-2PCT = 0.25 percent concentration
POPU	Total number of viable cells in the plating sample raised to some exponent printed directly below the abbreviation (i.e., EP + $6 = X \cdot 10^6$).
MUT 1	Total number of mutants or convertants obtained from the sample plated raised to some exponent printed directly below the abbreviation (i.e., EP + 0 = \times 100). For strain D4, MUT 1 represents the number of ADE+ convertants.
MUT 2	Only used for strain D4 and represents the number of TRY+ convertants in the plated sample.
FREQ 1	The calculated mutation or gene conversion frequency times the negative exponent written directly below. For strain D4, FREQ 1 represents the ADE+ value.
FREQ 2	Only used for strain D4 and represents the TRY+ conversion frequency.
CONTAM	Presence of contamination on any plates.
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DATA TABLE TERMS AND ABBREVIATIONS (continued)

ABBREVIATION OR TERM	DEFINITION OR EXPLANATION
AAF	2-Acetylaminofluorene
DMS0	Dimethylsulfoxide
DMN	Dimethylnitrosamine
EMS	Ethyl Methanesulfonate
QM	Quinacrine Mustard
NF	Nitrofluorene
SPECIES	Animal Strains
SPRDAW	Sprague Dawley Rats
ICRFLO	Flow ICR Random Bred Mice
RHESUS	Rhesus Monkey (<u>Macaca mulatta</u>)
MIXEDB	Dog, Mixed Breed

New Zealand White Rabbit

NEWZEA

LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM REPORT EXR34

COMPOUND FREQUENCY SUMMARY REPORT 04/25/75

SPECIES

COMPOUND 000299296

TEST	ORG	TA1535 HIS EX-8	TA1537 HIS EX-8	TA1538 HIS EX-8	0000D4 ADE EX-5	0000D4 TRY EX-5
NAN		1.60	5.71	4.89	2.89	46.67
NAP		1158.63	73.09	200.22	24.55	77.84
NA1	•	1.66	2.03	3.75	6.51	42.52
NA2		3.41	4.87	5.69	4.74	36.50

LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM REPORT EXR34

COMPOUND FREQUENCY SUMMARY REPORT 04/25/75

SPECIES ICRFLO COMPOUND 000299296

TEST	ORG.	TA1535 HIS EX-8	TA1537 HIS EX-8	TA1538 HIS EX-8	0000D4 ADE EX-5	0000D4 TRY EX-5
ACT	A+C	5.02	7.20	3.61	3.84	19.18
ACT	A+T	16.14	2.38	4.54	6.72	49.80
ACT	A-C	. 3.28	7.02	8.13	2.60	32.95
ACT	PLI	538.06	17.26	25.91	6.95	106.58
ACT	PLU	5.71	8.54	4.83	6.50	72.42
ACT	PTE	14.00	4.13	3.04	5.84	81.70
ACT	LI1	1.93	8.16	5.77	7.82	50.95
ACT	LI2	3.30	4.88	1.69	8.58	62.88
ACT	LU1	3.11	9.29	2.86	6.36	23.93
ACT	LU2	3.46	8.11	4.58	8.53	42.23
ACT	TEI	3.49	2.13	2.13	6.56	26.00
ACT	TE2	3.09	8.44	5.01	5.94	37.03

LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM REPORT EXR34

COMPOUND FREQUENCY SUMMARY REPORT 04/25/75

. SPECIES SPRDAW COMPOUND 000299296

TEST	ORG	TA1535 HIS EX-8	TA1537 HIS EX-8	HIS	TA1538 HIS EX-8	0000D4 ADE EX-5	000004 TRY EX-5
ACT	A+C	1.61	5.41		1.64	3.41	3.32
ACT	A+T		2.25		1.70	1.11	1.01
ACT	A-C	1.49	3.04	1.97	1.65	2.45	3.15
ACT	PLI	19.21	11.93		13.62	6.67	10.95
ACT	PLU	2.07	1.90		1.49	1.75	3.06
ACT	PTE	3.56	2.84		1.74	2.17	2.96
ACT	LII	1.94	3.86		2.83	2.69	2.98
ACT	L I 2	3.07	161.11	2.03	3.68	3.03	3.55
ACT	LUI	2.42	242.11	2.07	2.54	1.67	3.13
ACT	LU2	1.02	62.50	3.00	2.63	2.55	1.67
ACT	TE1	7.58	35.94	2.78	2.26	4.18	1.83
ACT	TE2	4.66	2.25		2.84	5.08	4.95

LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM REPORT EXR34

COMPOUND FREQUENCY SUMMARY REPORT 04/25/75

SPECIES RHESUS

COMPOUND 000299296

TEST	DRG	TA1535 HIS EX-8	TA1537 HIS EX-8	TA1538 HIS EX-8	TA1538 HIS EX-8	0000D4 ADE EX-5	0000D4 TRY EX-5
ACT	A+C	18.56	2.33	5.85		2.22	3.02
ACT	A+T	18.18	2.76	8.85		3.08	2.73
ACT	V -C	13.67	2.66	4.20	5.62	3.02	1.99
ACT	PL I	283.75	9.04	86.14	,,,,	5.98	5.82
ACT	PLU	12.85	2.33	4.58		2.41	
ACT	PTE	15.37	1.56	5.62		2.99	2.81
ACT	LII	19.13	2.51	152.94	43.75	3.17	3.95
ACT	L 1 2		3.26	47.87	66.04		4.95
ACT	LU1	11.28	2.43	16.10	00.04	3.04	4.80
ACT	LU2	18.35	2.06			5.09	4.95
ACT	TE1			10.06		3.15	3.72
		19.62	2.58	17.37		4.39	4.14
ACT	TE2	14.04	2.75	9.56		2.86	2 54

INTERPRETATION OF RESULTS AND CONCLUSIONS ٧.

Compound 000299296, Ferrous Gluconate, was evaluated for genetic activity in a series of in vitro microbial assays with and without metabolic activation. The following results were obtained:

- Α. Salmonella typhimurium
- Plate Tests 1.

At a concentration of 0.079%, 000299296 was not mutagenic for the bacterial indicator organisms used in the direct and activation plate tests.

2. Nonactivation suspension tests

The results of these tests were negative.

3. Activation suspension tests

The results of these tests indicate that this compound is positive in activation tests with TA-1538 and primate liver tissue. The tests were repeated and the results appeared positive. No other tests were positive with this compound.

- В. Saccharomyces cerevisiae
- 1. Nonactivation suspension tests

The results of these tests were negative.

2. Activation suspension tests

The results of these tests were negative.

С. Conclusions

Compound 000299296, Ferrous Gluconate, appeared to be mutagenic for indicator strain TA-1538 in activation tests with primate liver tissue. No other tests with this compound were positive.

Submitted by:

David Brusick, Ph.D. Director of Genetics



APPENDIX

Tabulation of Data



CONTRACT EXPERIMENT 502802 ORG			22374-2104 DETECTOR TA1535 SPECIES			PROJECT 02468 DATE - 04/25/75		
				POPU MUT1		FREQ1	74723713	
	COMPOUND	TEST ID	CONCENTRATION	EP+6	EP+0	EP-8	CONTAM	
		NAN	SALINE	0312	0005	. 1.60	0	
		NAP	EMS 0.002 %	0394	4565	1158.63	0	
	000299296	NAI	0007-2 PCT.	0421	0007	1.66	0	
	000299296	NA2	0035-3 PCT.	0528	0018	3-41		

CONTRACT EXPERIMENT 504102		22374-2104 DETECTOR TA1537	PROJECT 02468 SPECIES DATE - 04/25/75							
	CAP CRINGN,	J 0 4 1	<i></i>	DISTRICTION TAIDST SPECIES			S DATE - 04/25/75			
			ORG	·	POPU	MUT1	FREQ1			
1	COMPOUND	TEST	ID	CONCENTRATION	EP+6	EP+0	EP-8	CONTAM		
		NAN		SALINE	1435	0082	5.71	o		
		NAP		QM 1.0 UG/ML	1293	0945	73.09	0		
(000299296	NA1		0007-2 PCT.	1035	0021	2.03	1		
(000299296	NA2		0035-3 PCT.	1211	0059	4.87			

CONTRACT EXPERIMENT 507802		T 22374-2104 DETECTOR TA1538	SPE	CIES	PROJECT 02468 DATE - 04/25/75		
COMPOUND	ORG TEST ID	CONCENTRATION	POPU EP+6	MUT1 EP+0	FREQ1 EP-8	CONTAM	
	NAN	DMSO	0327	0016	4.89	0	
	NAP	NF 125 UG-ML	0465	0931	200.22	0	
000299296	NAI	0007-2 PCT.	0347	0013	3.75	0	
000299296	NA2	0035-3 PCT.	0334	0019	5.69	0	

REPORT EXR33 LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM COMPOUND SUMMARY BACKUP DETAIL

		CON	TRACT	22374-2104	PROJECT 02468						
EXPERIMENT 504104		DETECTOR 0000D4	SPECIES			DATE - 04/25/75					
			ORG		POPU	MUT1	MUT2	FREQ1	FREQ2		
	COMPOUND	TEST	ID	CONCENTRATION	EP+4	EP+1	EP+1	EP-5	EP-5	CONTAM	
		NAN		SALINE	0450	0013	0210	2.89	46.67	4	
		NAP		EMS 1.0 %	0501	0123	0390	24.55	47.84	6	
	000299296	NA1		0005-0 PCT.	0461	0030	0196	6.51	42.52	0	
	000299296	NA2		0025-1 PCT.	0548	0026	0200	4.74	36.50	0	

REPORT EXR33 LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM COMPOUND SUMMARY BACKUP DETAIL

CONTRACT			22374-2104	PROJECT 02468				
EXPERIMENT	5035	01	DETECTOR TA1535	SPE	CIES 1	CRFLO DATE -	- 04/25/75	
		ORG		POPU	MUT1	FREQ1		
COMPOUND	TEST	ID	CONCENTRATION	EP+6	EP+0	EP-8	CONTAM	
	A+C		DMN 50 UM/ML	0697	0035	5.02	O	
	A+T		***NO MATCH***	0694	0112	16.14	o	
	A-C		SALINE	0762	0025	3.28	0	
	ACP	LI	DMN 50 UM/ML	0762	4100	538.06	0	
	ACP	LU	DMN 50 UM/ML	0840	0048	5.71	o	
	ACP	TE	DMN 50 UM/ML	0750	0105	14.00	0	
000299296	ACT	LII	0007-2 PCT.	0726	0014	1.93	0	
000299296	ACT	LI2	0035-3 PCT.	0667	0022	3.30	0	
000299296	ACT	LU1	0007-2 PCT.	0805	0025	3.11	2	
000299296	ACT	LU2	0035-3 PCT.	0752	0026	3.46	0	
000299296	ACT	TE1	0007-2 PCT.	0774	0027	3.49	0	
000299296	ACT	TE2	0035-3 PCT.	0711	0022	3.09	0	

REPORT EXR33 LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM COMPOUND SUMMARY BACKUP DETAIL

			22374-2104	PROJECT 02468					
EXPERIMENT	T 5038	301	DETECTOR TA1537	SPE	CIES ICRFLO	DATE	- 04/25/75		
		ORG		POPU	MUT1	FREQ1			
COMPOUND	TEST	ID	CONCENTRATION	EP+6	EP+O	EP-8	CONTAM		
	A+C		AAF 800 UG/ML	1860	0134	7.20	. 0		
	A+T		***NO MATCH***	3195	0076	2.38	. 0		
	A-C		DMSO	1653	0116	7.02	o		
	ACP	LI	AAF 800 UG/ML	2126	0367	17.26	0		
	ACP	LU	AAF 800 UG/ML	0972	0083	8.54	2		
	ACP	TE	AAF 800 UG/ML	1452	0060	4.13	o		
000299296	AC T	LII	0007-2 PCT.	0944	0077	8.16	0		
000299296	ACT	LI2	0035-3 PCT.	1065	0052	4.88	2		
000299296	ACT	LU1	0007-2 PCT.	0915	0085	9.29	2		
000299296	ACT	LU2	0035-3 PCT.	0826	0067	8.11	2		
000299296	AC T	TE1	0007-2 PCT.	4601	0098	2.13	2		
000299296	ACT	TE2	0035-3 PCT.	0652	0055	8 • 44	2		

REPORT EXR33 LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM COMPOUND SUMMARY BACKUP DETAIL

	EXPERIMENT			T 22374-2104 DETECTOR TA1538 SPECIES I			PROJECT 02468 ICRFLO DATE - 04/25/75		
	CATERITIEN	2030	O1	DETECTOR TAISSO	376	CIES ICKE	LU DATE +	04/25/75	
			ORG		POPU	MUT1	FREQ1		
•	COMPOUND	TEST	ID	CONCENTRATION	EP+6	EP+0	EP-8	CONTAM	
		A+C		AAF 800 UG/ML	0859	0031	3.61	0	
		A+T		***NO MATCH***	1013	0046	4.54	o	
		A-C		DMSO	0541	0044	8.13	0	
		ACP	LI	AAF'800 UG/ML	0602	10156	25.91	0	
		ACP	LU	AAF 800 UG/ML	0766	0037	4.83	. 0	
		ACP	TE	AAF 800 UG/ML	1348	0041	3.04	2	
	000299296	ACT	LII	0007-2 PCT.	0659	0038	5.77	2	
	000299296	AC Ţ	LI2	0035-3 PCT.	2189	0037	1.69	0	
	000299296	ACT	LU1	0007-2 PCT.	1187	0034	2.86	2	
	000299296	ACT	LU2	0035-3 PCT.	0786	0036	4.58	2	
	000299296	ACT	TE1	0007-2 PCT.	1219	0026	2.13	0	
	000299296	ACT	TE2	0035-3 PCT.	0619	0031	5.01	o ·	

REPORT EXR33 LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM COMPOUND SUMMARY BACKUP DETAIL

CONTRACT			22374-2104			PROJECT 02468					
	EXPERIMENT	5037	01	DETECTOR 0000D4		SPECIES ICRFLO		ICRFLO	DATE - 04/25/75		
	40440		ORG			POPU	MUT1	MUT2	FREQ1	FREQ2	
	COMPOUND	TEST	ID	CONCENT	TRATION	EP+4	EP+1	EP+1	EP-5	EP-5	CONTAM
		A+C		DMN 90	UM/ML	0417	0016	0800	3.84	19.18	5
		A+T		***NO N	4A TC H***	0506	0034	0252	6.72	49.80	4
		A-C		SALINE		0346	0009	0114	2.60	32.95	0
		ACP	LI	DMN 90	UM/ML	0532	0037	0567	6.95	106.58	0
		ACP	LU	DMN 90	UM/ML	0446	0029	0323	6.50	72.42	6
		ACP	TE .	DMN 90	UM/ML	0377	0022	0308	5.84	81.70	0 .
	000299296	ACT	LII	0005-0	PCT.	0473	0037	0241	7.82	50.95	0
	000299296	A.C.T	LI2	0025-1	PCT.	0466	0040	0293	8.58	62.88	0
	000299296	ACT	LU1	0005-0	PCT.	0535	0034	0128.	6.36	23.93	0
	000299296	ACT	LU2	0025-1	PCT.	0457	0039	0193	8.53	42.23	0
	000299296	ACT	TE1	0005-0	PCT.	0427	0028	0111	6.56	26.00	4
	000299296	ACT	TE2	0025-1	PCT.	0505	0030	0187	5.94	37.03	4

REPORT EXR33 LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM COMPOUND SUMMARY BACKUP DETAIL

CONTRACT			22374-2104	PROJECT 02468				
EXPERIMEN	T 5028	301	DETECTOR TA1535	SPE	CIES SPRDAW		04/25/75	
COMPOUND	TCCT	ORG		POPU	MUT1	FREQ1		
COMPOUND	TEST	ΙD	CONCENTRATION	EP+6	EP+0	EP-8	CONTAM	
	A+C		DMN 50 UM/ML	0496	0008	1.61	o	
	A-C		SALINE	0941	0014	1.49	0	
	ACP	LI	DMN 50 UM/ML	0229	0044	19.21	2	
	ACP	LU	DMN 50 UM/ML	0242	0005	2.07	0	
	ACP	TE	DMN 50 UM/ML	0281	0010	3.56	2	
000299296	ACT	LII	0007-2 PCT.	0516	0010	1.94	0	
000299296	ACT	LI2	0035-3 PCT.	0326	0010	3.07	2	
000299296	AC T	LU1	0007-2 PCT.	0455	0011	2.42	o	
000299296	ACT	LU2	0035-3 PCT.	0491	0005	1.02	o	
000299296	ACT	TE1	0007-2 PCT.	0330	0025	7.58	2	
000299296	ACT	TE2	0035-3 PCT.	0279	0013	4.66	2	

REPORT EXR33 LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM

COMPOUND SUMMARY BACKUP DETAIL

CONTRAC			NTRACT	22374-21	PROJECT 02468					
	EXPERIMENT	5045	501	DETECTOR	TA1537	SPE	CIES S	SPRDAW	DATE - 04/25/75	1/25/75
			ORG			POPU	MUT1	FREQI		
	COMPOUND	TEST	ΙD	CONCENTRA	ATION	EP+6	EP+0	EP-8	CON	ITAM.
		A+C		AAF 800 I	UG/ML	1219	0066	5.41)
		A+T		***NO MA	TCH***	1245	0028	2.25	0)
		A-C		DMSO	•	1448	0044	3.04	. 0)
		ACP	LI	AAF 800 (JG/ML	0989	0118	11.93	0)
		ACP	LU	AAF 800 (UG/ML	1319	0025	1.90	0)
		ACP	TE	AAF 800 t	JG/ML	0952	0027	2.84	2	
	000299296	ACT	LI1	0007-2 PC	CT.	0856	0033	3.86	1	
	000299296	AC.T	LI2	0035-3 PC	т.	0018	0029	161.11	0	
	000299296	ACT	LU1	0007-2 PC	CT.	0019	0046	242.11	1	
	000299296	ACT	LUŻ	0035-3 PC	T.	0048	0030	62.50	3	
	000299296	ACT	TE1	0007-2 PC	CT.	0064	0023	35.94	3	
	000299296	ACT	TE2	0035-3 PC	т.	0932	0021	2.25	3	

CONTRACT 22374-2104					PROJECT 02468						
	EXPERIMENT 506210		10	DETECTOR TA1537	SPE	CIES SPRDAW	DATE	- 04/25/75			
	COMPOUND	TEST	ORG ID	CONCENTRATION	POPU EP+6	MUT1 EP+0	FREQ1 EP-8	CONTAM			
		A-C		DMSO	1523	0030	1.97	0			
	000299296	ACT	LI2	0035-3 PCT.	1775	0036	2.03	.2			
	000299296	ACT	LU1	0007-2 PCT.	1693	0035	2.07	. o			
	000299296	ACT	LU2	0035-3 PCT.	1566	0047	3.00	2			
	000299296	AC T	TE1	0007-2 PCT.	1834	0051	2.78	2			

CONTRACT		CT 22374-2104	PROJECT 02468					
EXPERIMENT	504301	DETECTOR TA1538	SPE	CIES SPRDA	W DATE -	04/25/75		
	ORO	;	POPU	MUT1	FREQ1			
COMPOUND	TEST ID	CONCENTRATION	EP+6	EP+0	EP-8	CONTAM		
	A+C	AAF 800 UG/ML	3528	0058	1.64	0		
	A+T	***NO MATCH***	4002	0068	1.70	o		
	A-C	DMSO	3028	0050	1.65	0		
	ACP L	AAF 800 UG/ML	2158	0294	13.62	0		
	ACP LU	J AAF 800 UG/ML	3414	0051	1.49	0		
	ACP TE	AAF 800 UG/ML	4136	0072	1.74	0		
000299296	ACT LII	0007-2 PCT.	1556	0044	2.83	0		
000299296	ACT LIZ	0035-3 PCT.	1495	0055	3.68	0		
000299296	ACT LU1	0007-2 PCT.	2006	0051	2.54	0		
000299296	ACT LUZ	0035-3 PCT.	1633	0043	2.63	0		
000299296	ACT TE1	0007-2 PCT.	1903	0043	2.26	0		
000299296	ACT TE2	0035-3 PCT.	1689	0048	2.84	. 0		

REPORT EXR33 LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM COMPOUND SUMMARY BACKUP DETAIL

CONTRACT EXPERIMENT 504401			11130131 02400									
	EXPERIMENT	5044	01	DETECTO	DR 0000D4	SPE	CIES S	PRDAW	DA	DATE - 04/25/75		
	COMPOUND	TEST	ORG ID	רחאר באי	TRATION	POPU EP+4	MUT1 EP+1	MUT2 EP+1	FREQ1	FREQ2	CONTAN	
		A+C	10	DMN 90		1085	0037	0036	EP-5 3.41	EP-5 3.32	CONTAM	
		A+T			1ATCH***	4064	0045	0041	1.11	1.01	0	
•		A-C		SALINE		1144	0028	0036	2.45	3.15		
		ACP	LI	DMN 90	UM/ML	1005	0067	0110	6.67	10.95	0.	
		ACP	LU	DMN 90	UM/ML	1142	0020	0035	1.75	3.06	0	
		ACP	TE	DMN 90	UM/ML	1150	0025	0034	2.17	2.96	0	
	000299296	ACT	LI1	0005-0	PCT.	1042	0028	0031	2.69	2.98	0	
	000299296	ACT.	LI2	0025-1	PCT.	0957	0029	0034	3.03	3.55	0	
	000299296	ACT	LU1	0005-0	PCT.	1021	0017	0032	1.67	3.13	o	
	000299296	ACT	LU2	0025-1	PCT.	1137	0029	0019	2.55	1.67	0	
	000299296	ACT	TE1	0005-0	PCT.	0981	0041	0018	4.18	1.83	0	
	000299296	ACT	TE2	0025-1	PCT.	0768	0039	0038	5.08	4.95	0	

REPORT EXR33 LITTON BIONETICS MUTAGENIC ACTIVITY SYSTEM COMPOUND SUMMARY BACKUP DETAIL

EXPERIMENT		CONTRACT 504901		22374-2104 DETECTOR TA1535		SPE	PROJECT 02468 SPECIES RHESUS DATE - 04/2				
			ORG			POPU	MUT1				
COMPO	UND	TEST	ID	CONCEN	TRATION	EP+6	EP+C	· = - -	CONTAM		
		A+C		DMN 50	UM/ML	1546	0287	18.56	0		
		A + T		***N()	MATCH***	2718	0494	18.18	0		
		A-C		SALINE	•	2187	0299	13.67	o		
		ACP	LI	DMN 50	UM/ML	2080	5902	283.75	0		
		ACP	LU	DMN 50	UM/ML	1666	0214	12.85	0		
		ACP	TE	DMN 50	UM/ML	1971	0303	15.37	o		
00029	9296	ACT	LII	0007-2	PCT.	1636	0313	19.13	0		
00029	9296	ACT	L I 2	0035-3	PCT.	1897	0500	26.36	0		
00029	9296	ACT	LUI	0007-2	PCT.	2251	0254	11.28	o		
00029	9296	ACT	LU2	0035-3	PCT.	1646	0302	18.35	0		
00029	9296	ACT	TEI	0007-2	PCT.	1478	0290	19•62	0		
00029	9296	ACT	TE2	0035-3	PCT.	1859	0261	14.04	2		

				22374-2104	PROJECT 02468					
EXPERIMENT 505001		DETECTOR TA1537	SPE	CIES RHESUS	DATE -	DATE - 04/25/75				
	COMPOUND	TEST	ORG ID	CONCENTRATION	POPU EP+6	MUT1 EP+0	FREQ1 EP-8	CONTAM		
		A+C		AAF 800 UG/ML	2665	0062	2•33	0		
		Δ+T		***NO MATCH***	3079	0085	2.76	0		
		A-C		DMSO	2597	0069	2.66	0		
		ACP	LI	AAF 800 UG/ML	2899	0262	9.04	0		
		ACP	LU	AAF 800 UG/ML	2577	0060	2.33	0		
		ACP	TE.	AAF 800 UG/ML	3459	0054	1.56	2		
	000299296	ACT	LII	0007-2 PCT.	2710	0068	2.51	0		
	000299296	ACT.	LI2	0035-3 PCT.	1992	0065	3.26	0		
	000299296	ACT	LU1	0007-2 PCT.	2305	0056	2.43	0		
	000299296	AC T	LU2	0035-3 PCT.	2378	0049	2.06	0		
	000299296	ACT	TE1	0007-2 PCT.	2283	0059	2.58	2		
	000299296	ACT	TE2	0035-3 PCT.	2511	0069	2.75	2		

CONTRACT			TRACT	22374-210	PROJECT 02468					
EXPERIMENT		5051	01	DETECTOR TA1538		SPE	CIES	DATE - 0	04/25/75	
			ORG		•	POPU	MUT1	FREQ1		
	COMPOUND	TEST	ID	CONCENTRA	MOITA	EP+6	EP+0	EP-8		CONTAM
		A+C		AAF 800 U	JG/ML	0855	0050	5.85		0
		A+T		***NO MA1	TCH***	0599	0053	8.85		0
		, A -C		DMSO		0834	0035	4.20		. 0
	<i>,</i>	ACP	LI	AAF 800 t	JG/ML	0866	0746	86.14	· .	0
		ACP	LU	AAF 800 U	IG/ML	0896	0041	4.58	•	0
		ACP	TE .	AAF 800 L	JG/ML	0836	0047	5.62		O
	000299296	ACT	LII	0007-2 PC	Τ.	0034	0052	152.94		0
	000299296	ACT	LI2	0035-3 PC	т.	0094	0045	47.87		0
	000299296	AC T	LU1	0007-2 PC	Т.	0205	0033	16.10		о .
	000299296	ACT	LU2	0035-3 PC	т.	0328	0033	10.06		0
	000299296	AC T	TE1	0007-2 PC	T.	0213	0037	17.37		0
	000299296	ACT	TE2	0035-3 PC	т.	0251	0024	9.56		. 0

				22374-2104	PROJECT 02468					
EXPERIMENT 506209			09	DETECTOR TA1538	SPECIES RHESUS DATE			- 04/25/75		
			ORG		POPU	MUT1	FREQ1			
	COMPOUND	TEST	ID	CONCENTRATION	EP+6	EP+0	EP-8	CONTAM		
		V-C		DMSO	0427	0024	5.62	. 0		
	000299296	ACT	LII	0007-2 PCT.	0080	0035	43.75	o		
	000299296	ACT	LI2	0035-3 PCT.	0053	0035	66-04	. 0		

CONTRACT			22374-21	104	PROJECT 02468						
EXPERIMENT 504802		DETECTOR 0000D4		SPECIES RHESUS			DATE - 04/25/75				
			ORG			POPU	MUT1	MUT2	FREQ1	FREQ2	
	CUMPOUND	TEST	ID	CONCENTR	RATION	EP+4	EP+1	EP+1	EP-5	EP-5	CONTAM
		A+C		DMN 90 L	JM/ML	1127	0025	0034	2.22	3.02	0
		A+T		***NO MA	1 TCH***	1427	0044	0039	3.08	2.73	0
		A-C		SALINE		1257	0038	0025	3.02	1.99	0
		ACP	ĻΙ	DMN 90 U	JM/ML	1322	0079	0077	5.98	5.82	0
		ACP	LU	DMN 90 (JM/ML	1493	0036	0042	2.41	2.81	0
		ACP	TE	DMN 90 L	JM/MĹ	1139	0034	0045	2.99	3.95	0 .
	000299296	ACT	LII	0005-0 F	PCT.	1232	0039	0061	3.17	4.95	4
	000299296	AC.T	LI2	0025-1 F	PCT.	1021	0031	0049	3.04	4.80	. 4
	000299296	ACT	LUI	0005-0 F	PCT.	0727	0037	0036	5.09	4.95	4
	000299296	ACT	LU2	0025-1 F	PCT.	1047	0033	0039	3.15	3.72	0
	000299296	ACT	TE1	0005-0 F	PCT.	0797	0035	0033	4.39	4.14	4
	000299296	ACT	TE2	0025-1 F	PCT.	0978	0028	0025	2.86	2.56	0